AF&PA

AMERICAN FOREST & PAPER ASSOCIATION
Regulatory Affairs

November 23, 1998

TO: NTP
Report on Carcinogens Subcommittee

RE: Dioxin Re-evaluation

Please find enclosed comments on the re-evaluation of dioxin, submitted on behalf of the members of the American Forest & Paper Association.

Our filing includes a very short *Executive Summary*, followed by sections addressing the human evidence and the mechanistic evidence regarding dioxin's potential carcinogenicity. Our review of the human evidence focuses on publications subsequent to the IARC review that are included in NTP's supplemental Background Document. In addition, we include fuller critiques of two of the recent human studies. The mechanism discussion highlights the uncertainty regarding the precise mechanism of carcinogenicity, and includes a letter to NTP from a number of distinguished mechanism researchers.

In your evaluation of dioxin, we urge that you take into account the key information covered in our submission:

(1) In 1997, the International Agency for Research on Cancer (IARC), concluded that the evidence for the carcinogenicity of dioxin in humans is "limited," based on small excesses of overall cancer risk. Under IARC's definition, this means that with respect to the human data, it is not possible to rule out chance, bias or confounding in the cancer studies evaluated. Since IARC's evaluation, no new studies have been published that would alter that determination. In fact, recent updates of the Seveso and Ranch Hand cohorts show no statistically significant excesses in overall cancer.

(2) Despite the limited human evidence, IARC placed dioxin in its Group I category by invoking mechanistic considerations based on the Ah receptor model. However, as indicated in the attached submission on mechanistic data prepared by Dr. Tom Starr, we are still a long way from understanding how dioxin may or may not cause cancer in rodents, much less humans. Ah receptor binding is not in and

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of itself sufficient, and we do not presently know the mechanism of carcinogenicity. IARC itself acknowledges this fact in stating that "even though Ah receptor activation is likely to be required for the carcinogenicity of 2,3,7,8-TCDD, its precise role in this process remains unclear." (Monograph at p. 331.)

As indicated in a letter attached at Tab B-1, eminent dioxin and cancer researchers, including the discoverer of the Ah receptor, strongly disagree that there is sufficient understanding of the complex molecular events to conclude on mechanistic grounds that dioxin in fact will cause cancer in humans.

Thus, the mechanistic data are not an adequate basis for elevating the "limited" human evidence to classify dioxin as a known human carcinogen.

It is incumbent on NTP as a scientific agency to base its classifications on factual scientific evidence, and not on unproven hypotheses. Given the limited human data, and the incomplete understanding of the mechanism of carcinogenicity, there is not an adequate basis for designating dioxin a known human carcinogen. We urge you to reject such a classification.

Sincerely,

John L. Festa, Ph.D.
Senior Scientist

Enclosure

COMMENTS OF THE AMERICAN FOREST AND PAPER ASSOCIATION ON THE PROPOSED NTP RECLASSIFICATION OF DIOXIN

November 23, 1998

EXECUTIVE SUMMARY

NTP's proposal to elevate 2,3,7,8-tetrachlorodibenzo-*para*-dioxin (TCDD or dioxin) to a "known to be a human carcinogen" category in the *Report on Carcinogens* is not supported by sufficient evidence of a causal connection with cancer in humans, nor is it warranted based on mechanistic evidence.

- There is no basis for an assessment of the human data different from IARC's classification of the evidence as *"limited,"* even considering post-IARC publications.

- There is reason, however, to differ with IARC's use of Ah receptor binding mechanism data to justify a "known human carcinogen" classification. Ah receptor binding is necessary but not sufficient for carcinogenesis, and the subsequent steps in potential carcinogenesis are still highly speculative. The available mechanistic data thus are not adequate basis for elevating the classification of dioxin to a known human carcinogen.

Human Carcinogenicity Data

IARC's Working Group on dioxin convened in February 1997, and concluded that there is *"limited evidence* in humans" for the carcinogenicity of dioxin. (IARC Monograph at 342.) IARC's conclusion was based on slightly elevated risk for all cancers combined. The possibility of confounding could not be excluded, and the fact that the excess risk is limited to all cancers combined without predominant sites means that the findings must be treated with caution. (Monograph at 347.)

The proposed NTP listing language (1997 RC Draft Background Document at page RC-1) cites only an "association" with certain cancers and all cancers combined -- far short of the criterion of sufficient evidence of a causal relationship. The proposed listing language also cites a Seveso study as new; yet, IARC considered data from the 15-year followup of Seveso, and the study's author, Dr. Bertazzi, was a member of the Working Group. Moreover, the analysis in the recent *Bertazzi* publication does not

"further strengthen the association" that IARC evaluated; rather, the data show no statistically significant excess for all cancers combined, and thus weakens the association.

In 1998, NTP offers a number of "selected epidemiologic reports" as a supplement to the 1997 RC Draft Background Document. The human evidence offered in the 1998 supplement is summarized at Tab A. Critiques of the *Bertazzi* and *Hooiveld* data by Dr. Raymond Greenberg are presented at Tabs A-1 and A-2.

The ostensibly new publications in the 1998 supplement do not present any evidence that would justify a different conclusion than the one reached by IARC with respect to the human data. Indeed, the essential data from all but one publication (which NTP notes that the authors acknowledge to be unreliable in its assessment of exposure from PCB transformer oil and herbicides in soil) were considered by IARC in its 1997 proceedings. In short, no new developments have occurred that warrant a conclusion different from the conclusion reached by IARC -- that the human evidence is *limited*.

Mechanistic Data

Available mechanistic data simply do not rise to the level necessary to support classification of dioxin as a known human carcinogen. While much is known about Ah receptor binding, receptor binding is not sufficient to induce cancer. Many hypotheses regarding the precise mechanism of carcinogenicity have been put forward in the scientific literature, but none has been established as scientific fact:

- Induction of enzymes such as CYP1A1 following receptor binding is well established, but it is not known whether this mechanism is relevant to the process of carcinogenesis.

- While cell proliferation is widely believed to be a necessary step for carcinogenesis, the role of Ah receptor binding in increased cell proliferation is unclear. For example, *Walker (1998)* found that dioxin concentrations in the rat liver did not correlate with the cell proliferation response.

- Mechanisms other than Ah receptor binding have been reported for some of TCDD's effects. *See, e.g., Enan (1998)*(guinea pig toxicity too rapid to involve gene expression subsequent to receptor occupancy); *Hossain (1998)*(dioxin induced apoptosis in human leukemic T cells that lack Ah receptor). As *Hossain, et al.*, concluded, "not all the biological effects of TCDD can be explained using this receptor-based model."

As more fully addressed at Tab B, recent mechanistic research has not resolved the many substantial uncertainties surrounding the numerous possible sequelae of Ah receptor binding. As of now, we cannot identify with any certainty the specific mechanistic steps or events that contribute to the carcinogenicity of dioxin in rodents. In fact, the IARC monograph is replete with multiple explanations of the potential mechanism of carcinogenicity of dioxin. Factors and processes other than receptor binding may well provide the critical determinants of whether dioxin is carcinogenic to humans.

Indeed, while much is known about the Ah receptor binding mechanism, receptor binding is only the first step in a "multiple pathway web of interactions that may, or may not, in any given situation, lead to a toxic response." This characterization comes from comments on NTP's proposal by a group of distinguished academic mechanistic researchers including Dr. Al Poland, one of the pioneers in dioxin research and now at CDC/NIOSH; Dr. Christopher Bradfield of McArdle Labs at the University of Wisconsin, Dr. William Greenlee and Vice Chancellor Edward Bresnick of the University of Massachusetts, and Dr. Thomas Sutter of Johns Hopkins. As these scientists state in their comments to NTP:

> Lack of understanding of the complex molecular events downstream from receptor occupancy that *might or might not culminate in malignancy* prevents one from concluding on mechanistic grounds that TCDD is a *Known Human Carcinogen* at the present time.

Letter to NTP, Tab B-1 (emphasis added).

Clearly the mechanistic evidence does not justify upgrading the classification of dioxin to known human carcinogen status at this time. Without knowledge of dioxin's carcinogenic mechanism in rodents, one cannot confidently conclude that dioxin is known to be carcinogenic in humans. The mechanistic data regarding dioxin carcinogenicity remain insufficient to categorically declare this compound to be a human carcinogen.

HUMAN CARCINOGENICITY DATA

There Is No Basis for an Assessment Different from IARC's Classification of the Human Evidence as "Limited"

IARC's Working Group on dioxin convened in February 1997, and concluded that there is *"limited evidence* in humans for the carcinogenicity of 2,3,7-8 tetrachloro-dibenzo-para-dioxin." (IARC Monograph at 342.)[1]

None of the ostensibly new publications -- the "selected epidemiologic reports" provided by NTP as a supplement to the 1997 Background Document -- presents any evidence that would justify a different conclusion. The essential data from all but one publication (which NTP notes is acknowledged to be unreliable in its assessment of exposure from PCBs and herbicides in soil) were considered by IARC in its 1997 proceedings.

A. IARC's Evaluation

IARC placed the greatest emphasis on four occupational cohorts of herbicide producers who had both the highest exposure and the longest latency of the groups studied. In subcohorts with the longest latency and highest exposure within the cohorts, the risk for all cancers combined was increased approximately 1.4-fold. (Monograph at 337.) However, the possibility of confounding cannot be excluded, and the fact that the excess risk is limited to all cancers rather than a particular site means that the findings must be treated with caution. (Id.)[2]

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[1] IARC Working Groups classify human evidence as *limited* when:

> A positive association has been observed between exposure to the agent, mixture, or exposure circumstance and cancer for which a causal interpretation is considered by the Working Group to be credible, but chance, bias or confounding could not be ruled out with reasonable confidence.

[2] Indeed, we note that there are a number of significant limitations of the human evidence, including:

- The potential effects of dioxin and those of the products in which it was found could not separated from one another in all but a few of the epidemiological studies.

(continued...)

B. Epidemiologic Evidence Provided by NTP Other than IARC's Evaluation

1. September 1997 Documentation

The 1997 Draft NTP Background Document proposes text for the RC listing which states with respect to the human evidence:

"human studies have found an association between dioxin exposure and cancer mortality with respect to all cancers combined, non-Hodgkin's lymphoma, and lung cancer." (at p. RC-1)

Note that this statement merely goes to association, not causation. The 1997 RC proposed listing language further notes that IARC did not consider *Bertazzi* (1997, then in press). As discussed below, IARC did in fact consider the Bertazzi data, which -- contrary to NTP's assertion -- does not strengthen the association between dioxin exposure and human cancer.

Bertazzi (1997)

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[2](...continued)

- The magnitudes of all of the reported cancer risks are low or highly uncertain due to small case numbers, leaving the excess risks that might be attributable to dioxin exposure highly susceptible to the influences of chance (especially when the problem of multiple comparisons is taken into account for site-specific cancers), bias, and/or confounding.

- Smoking and other carcinogenic exposures cannot be confidently ruled out as causes of at least part of the reported excesses in mortality from all cancer, lung cancer, or the other inconsistent site-specific findings that have been noted in some studies. Adjustments for occupational, lifestyle-specific, and other confounding exposures have not generally been undertaken, and, in the few cases where they have been attempted, the treatment of confounding was inadequate.

- There is a pronounced lack of consistency among the studies in the site-specific cancer excesses (e.g., soft tissue sarcoma, non-Hodgkin lymphoma, digestive system cancers, and multiple myeloma) that have been reported.

NTP distributed the Bertazzi manuscript ("Dioxin Exposure and Cancer Risk: A 15-Year Mortality Study after the Seveso Accident") along with the draft Background Document in 1997. This Bertazzi paper, published in November 1997, does not add significantly to the data already available to the IARC Working Group in February of 1997. IARC had the 15-year followup data from Seveso, and Dr. Bertazzi was a member of the IARC Working Group. See IARC Monograph at page 161; see also Monograph Table 33, pp. 146-47. While the data are tabulated differently in the published "15-Year Mortality" paper, there are no statistically significant increases in all cancers reported in the published paper. See Tab 1 for a critique of the Seveso data by Dr. Raymond Greenberg, previously submitted to NTP.

2. 1998 NTP Supplemental Documentation

NTP this year has provided a supplemental report with one-page summary of "selected epidemiologic reports" published after the September 1997 Background Document, accompanied by copies of a number of ostensibly new studies. *The materials contained in the package are not news*.

In addition to *Bertazzi (1997)*, noted above, the 1998 package addresses the following:

Bertazzi (1998) - which is a review paper based on a 1996 presentation at a dioxin symposium. The review paper presents no new data.

Kogevinas 1997 - This June 1997 publication on the IARC international cohort was considered when the Working Group met in February of 1997. The study was discussed in detail at page 160 of the Monograph. See also page 144 for the tabular summary, and the reference for the *in press* manuscript at page 570. The *Kogevinas* study combines 36 cohorts from 12 countries. Some of these cohorts have been analyzed individually and the results published. Even in the combined analysis, there was only a small increase in all cancers (SMR 1.12), and no consistent dose-response relationship was noted.

Hooiveld (1998) - The results of the *Hooiveld* cohort were included in the *Kogevinas* data considered by IARC. (*Hooiveld* at page 892). Hooiveld reports a relative risk of 4.8 at medium exposure (defined as 7.7 to 124.1 ppt), and a relative risk of 4.4 at high exposure (defined as the rather wide range of 124.2 to 7,037 ppt) -- a decrease in relative risk despite the dramatic difference in exposure at the higher levels. Thus, this study does not show a dose-response relationship or causal connection. See Tab 2 for Dr.

Greenberg's previously submitted critique of the *Hooiveld* paper. The NTP summary does not note the results of the study.

Flesch-Janys (1998) - IARC reviewed publications by Flesch-Janys on a German cohort studied earlier by Manz. The IARC evaluation covered data from 1995 and 1996 publications in detail. For example, see the Monograph at page 155. The 1998 publication cited in the NTP supplement to the Background Document publishes information presented at a conference in Heidelberg in 1996 prior to the IARC meeting. According to NTP's summary in the supplement to the Background Document, the 1998 publication "employed new methods to estimate dose rates of TCDD." However, both this paper and the papers reviewed by IARC were based on followup of the cohort through 1992.

Lynge (1998) - This Danish study of workers manufacturing non-dioxin contaminated herbicides was part of the *Kogevinas* international cohort considered by IARC. The *Lynge* data were presented at the 1996 Heidelberg conference prior to the IARC meeting, though not published until 1998.

Becher (1998) - is a quantitative risk assessment using the *Flesh-Janys* data, which does not provide new data or SMR analysis. The Becher paper is also based on a presentation at the same 1996 conference as the Flesch-Janys paper, prior to IARC's meeting.

Michalek (1998) - Michalek is the published account of the most recent update of the Air Force Ranch Hands study, tracking mortality through 1993. IARC reviewed the Air Force's Interim Technical Report on Mortality through 1993 (cited in the Monograph at pages 168, 171, and 568 as *Ketchum & Akhtar* (1996)). The NTP summary cites the study without mention of the fact that it shows no increase in all cancers (SMR 0.9).

Hay and Tarrell (1997) - NTP's final citation was indeed not considered by IARC. As NTP notes, the authors acknowledge that their estimation of exposure to utility workers from PCB transformer oil and herbicides using soil levels is unreliable. SMRs for all cancers were generally not statistically significant, and there was no dose-response analysis.

In sum, nothing in the 1997 or 1998 packages provides any basis for reaching a different evaluation of the human data from that of the IARC Working Group, which classified the human evidence as *"limited."* The limited human evidence does not

support an upgrade in the classification of TCDD to *Known Human Carcinogen* status in the 9th *Report on Carcinogens*.

October 27, 1997
VIA Facsimile: 919/541-0295

NTP Board of Scientific Counselors' Report on Carcinogens Subcommittee
c/o Dr. Larry G. Hart, Executive Secretary
NIEHS
Research Triangle Park NC

Re: RC Draft Background Document for TCDD

Dear Members of the Subcommittee:

As a cancer epidemiologist and an invited observer to the IARC working group evaluation of the carcinogenicity of dioxin, I have considerable interest in the resulting report and the use of that report by other organizations. It is in that context that I have been asked by the American Forest and Paper Association to review the document referenced above, as well as the "new" study on the Seveso cohort that was distributed with the document. You will find attached a copy of my comments on the draft document and the updated report on mortality from the Seveso cohort.

I hope that you find these comments to be helpful in your deliberations. In addition, I plan to attend the public hearing on this topic later this week and would be grateful for the opportunity to make a few remarks to the Subcommittee at that time. Thanks in advance for your consideration.

Sincerely,

Raymond S. Greenberg, MD, PhD
Charleston SC

Enclosures

Comments on:
Bertazzi et al., Dioxin Exposure and Cancer Risk.
***Epidemiol*, (in press)**

Raymond S. Greenberg, MD, PhD
Charleston SC

- **In this paper, a large number of potential cancer outcomes were examined, few of which revealed positive associations.**

  This paper included analyses that presented separate results stratified simultaneously by cancer type, zone (of exposure) and gender. There were 27 types of cancer for males (including all cancers combined) and 26 types of cancer for females. In addition, there were three exposure zones. Accordingly, there were (27 x 3) + (26 x 3) = 159 reported outcomes. If these were all independent, and dioxin exposure was not related to cancer mortality, one would still expect 8 results to be statistically significant at the 0.05 level (159 x .05). In fact, a total of 8 results were observed to be statistically significant at the 0.05 level. Accordingly, the findings reported here are entirely consistent with chance.

- **There was a lack of internal consistency in most of the reported findings, which was unlikely to have arisen from biological differences.**

  Most of the positive associations observed among males were not found among women and *vice versa*. For example, the authors cited an excess of rectal cancer deaths among males in zone B, with virtually no association seen among women. Among males in zone R, an excess of respiratory cancer deaths was reported for males, but there was an apparent deficit among females. An excess of deaths from leukemia among men was reported for zone B, but fewer than expected deaths were found for women. In zone B, women were reported to have an excess of deaths from multiple myeloma, but no such excess was found for males. In zone R, there was an apparent excess of deaths from soft tissue sarcomas among men, but no soft tissue sarcoma deaths were reported among women (with 1.5 expected).

  Although it is possible to speculate that biological differences between males and females might explain gender differences in the risks of certain cancers, such explanations seem unlikely for the particular cancer sites. None of the cancers in question is known or even suspected to be sex hormone-dependent. Interestingly, cancers that are known to be sex hormone-related, such as those involving the breast, ovaries, uterus, and prostate, did not reveal any excesses in mortality in this investigation. It is unlikely, therefore, that the gender differences observed in this study are biologically based. It is

more likely that the inconsistencies reflect either false-positive associations where the excesses were noted, or false negative results where associations were not observed. The likelihood of obtaining false negative results in a study is measured by the statistical power, which is directly related to the sample size, the frequency of the outcome and the magnitude of any true effect. For zones B and R, the present study had a sample size that was sufficiently large to detect moderate to strong associations for all cancers combined and for lung cancer. In that regard, it is worth noting that for both men and women, no excess was observed in either zone B or R for these outcomes. While one cannot rule out a very weak association from these results, these data do not support an association at the strength typically observed with cause-and-effect relationships (a relative risk of two or three).

- **The isolated positive associations did not reveal any clear dose-response relationship.**

  The most heavily exposed population was that residing in zone A. For males, there was a borderline statistically significant **deficit** of all cancer deaths combined, with less than half the number expected. Summing across gender, the observed number of all cancer deaths combined was 16, with an expectation of 23. For digestive cancer, one of those highlighted by the authors, the number of deaths among males in zone A was one-fifth of the expectation, a result that approached statistical significance. For soft tissue sarcomas, a cancer that has been associated with dioxin exposure in other settings, there was no evidence of any excess in either of the two most heavily exposed zones. Nevertheless, the authors chose to highlight in their abstract the small, non-statistically significant excess mortality that was observed only among males in zone R, the least contaminated area.

- **The findings were largely inconsistent with those from other studies on human populations heavily exposed to dioxin.**

  In its summary of the evidence concerning the potential carcinogenicity of dioxin among humans, the International Agency for Research on Cancer (IARC) working group found that the most consistent positive association was for all cancers combined. Less convincing evidence was found for site-specific cancers, such as lung cancer, non-Hodgkin lymphoma, and soft-tissue sarcoma. In contrast, the Bertazzi et al paper reported no excess of deaths from all cancers combined. Summing results across all three zones and both genders, the observed total number of cancer deaths was 1,176, with an expectation of 1,259.7 (Relative Risk = RR = 0.93). The total number of lung cancer deaths was 245, as compared with an expectation of 257.9 (RR = 0.95). For non-Hodgkin lymphoma, the observed total number of deaths was 20, with 21.7 expected (RR = 0.92). For soft tissue sarcoma, the

observed number of deaths was 4, with 3.97 expected (RR = 1).

- **The authors attempted to tie together disparate findings by aggregating them into broad organ system categories (e.g., digestive diseases), which ignore basic etiologic dissimilarities.**

  The authors refer to excesses of "digestive" cancers among women in zone A and after ten years of latency in zone B. For zone B, the authors also report more specific excesses of stomach cancer mortality among women and rectal cancer among men. The category of digestive cancers includes malignancies of any of the following organ sites: esophagus, stomach, colon, rectum, hepatobiliary tract, liver, pancreas, and other unspecified sites. Epidemiologically, cancers of these anatomic sites are quite distinct. For example, beverage alcohol consumption appears to be a risk factor for esophageal cancer and perhaps rectal and pancreatic cancer, but not for the other sites. Similarly, cigarette smoking is a risk factor for esophageal and stomach cancers, but not for the other sites. Infection by *Helicobacter pylori* appears to play an important role in the development of stomach cancer, but not in the other sites. All of the known epidemiology suggests that these anatomic sites have different and distinct risk factor profiles, and therefore, the usual and customary manner of investigating them is to treat them as unique outcomes. Linking isolated findings for different anatomic sites into a broad and ill-defined aggregate category may falsely imply consistency of results, but it makes little sense from an etiologic point of view.

- **The results of the latency analysis are inconsistent and only selectively presented.**

  For all cancers combined, the risk of death appeared to increase beyond 10 years among women, but not for men. Similarly, the mortality from "digestive" cancers appeared to increase after 10 years among women, but not among men. In fact, the only cancer site presented for which there was an increased risk of death with increased latency among men was rectal cancer, and no results were presented for this site for women. The authors presented latency analyses only for zone B and only for selected cancer sites. No statistics were presented to help judge whether or not chance was a likely explanation of the observed latency results. For most of the site-specific analyses presented, the numbers of observed deaths were small, and therefore, the resulting estimates of effect were highly unstable (as reflected by broad confidence intervals).

- **Alternative explanations for reported outcomes are not thoroughly explored.**

  For males in zone B, three pleural cancers were observed, with an

expectation of 0.6. An obvious question is whether these individuals had any exposure to asbestos, which is the principal known cause of mesothelioma. For such a small number of affected individuals, inquiry could have been made into their occupational histories at minimal effort and expense, and such information would have proven valuable in the interpretation of the present findings.

Comments on: RC Draft Background Document for TCDD

Raymond S. Greenberg, MD, PhD
Charleston SC

- **Pg. RC-1, lines 17-18: "In the highly exposed industrial sub-cohorts, a causal relationship between TCDD exposure and mortality form all cancers combined was noted. . ."**

  The report of the IARC Working Group did not characterize the association as *causal.* In fact, the report states clearly that "While this relative risk does not appear likely to be explained by confounding, this possibility cannot be excluded" (p. 337). The report goes on to add that: "This lack of precedent for a multi-site carcinogen without particular sites predominating means that the epidemiological findings must be treated with caution" (p. 337). Finally, the report concluded that "there is *limited evidence* in humans for the carcinogenicity " of TCDD (p. 342). Thus, the association with all cancers combined was not deemed to be sufficient for causal inference.

- **Pg. RC-1, lines 19-21: "Increased risk for certain cancers was also reported in a new study of the Seveso, Italy, dioxin-exposed population (Bertazzi et al., 1998 [in press])."**

  The paper in question is not from a *new* study. Rather, as indicated subsequently in the document (page 3-1), this is an update of a cohort that has been followed since 1976, and about which reports have appeared multiple times in the peer-reviewed literature (see Bertazzi et al., *Am. J. Epidemiol,* 1989; **129**:1187-99, Bertazzi et al., *Organohalogen Compounds* 1996; **30**:294-6).

- **Pg RC-1, lines 21-23: "The additional findings were not considered in the IARC evaluation and further strengthen the association between dioxin-exposure and human cancer."**

  The 15-year follow-up results of Bertazzi et al., 1998 (in press) were available to the IARC Working Group. They were presented at an international dioxin meeting and reported in a peer-reviewed publication (Bertazzi et al., *Organohalogen Compounds* 1996; **30**:294-6). These findings are summarized in Table 33 of the IARC report (pp. 146-147) and in the corresponding text (p. 161). The only aspect of the new paper that was not cited in the IARC report was the subanalysis of the results by latency period. Because these latency results were not yet accepted by a peer-reviewed journal, the IARC Working Group felt that it was inappropriate to

include them in its report, and therefore, did not include the Seveso cohort in summary Table 38.

- **Pg. 3-2, lines 2-3: "In the largest and most heavily exposed German cohort, a dose-response relationship was noted for overall cancer mortality."**

  The dose-response relationship in question can be found in Table 39 (p. 194) of the IARC report. As noted in the IARC report: "This is largely the result of the high RR for the highest exposure group." Since there was not a smooth graded increase across the full range of exposure levels studied, it is inappropriate to use a linear test for trend, as was done by the authors of the paper in question. The test statistic resulting from a linear test for trend does not represent a valid estimate of the statistical significance of the observed pattern. The entire relationship was driven by a single outlying observation point. Moreover, only about 15 percent of this cohort had actual dioxin measurements; the doses of the remainder were imputed from job category and years of employment. Finally, the citation for this dose-response trend comes from an erratum, which is hardly reassuring about such a heavily emphasized finding.

- **Pg. 3-2, lines 6-8: "In its summary of the epidemiological evidence for the most highly exposed populations, the IARC Working Group identified a causal association between TCDD exposure and all cancer combined . . ."**

  As noted above, the IARC Working Group did not conclude that the epidemiological evidence supported a *causal* interpretation for the all cancer combined results. The report clearly concluded that confounding could not be excluded as a possible explanation (p. 337), and further suggested caution in any inference of causality given the absence of any strong site-specific associations. The IARC Working Group therefore concluded that there was *limited* (as opposed to sufficient) evidence in humans for carcinogenicity.

67 Legare Street, #403
Charleston, SC 29401
June 9, 1998

Dr. C.W. Jameson
National Toxicology Program
Report on Carcinogens
79 Alexander Drive, Building 4401
P.O. Box 12233
Research Triangle Park, NC 27709

Dear Dr. Jameson:

As a cancer epidemiologist and an invited observer at the February 1997 IARC working group evaluation of the potential human carcinogenicity of dioxin-like compounds, I have considerable interest in the resulting report and the use of that report by other organizations. It was in that context and on behalf of the American Forest and Paper Association that I submitted written comments on the *RC Draft Background Document for TCDD.* I also provided a critique of the report by Bertazzi *et al.*, entitled *Dioxin Exposure and Cancer Risk*, for consideration by members of the NTP Board of Scientific Counselors' Report on Carcinogens Subcommittee at their 30-31 October 1997 meeting.

At that meeting, I discussed briefly my reservations about the proposal to list TCDD as a *known human carcinogen.* During his presentation of the evidence in support of that proposal, Dr. Arnold Schecter made reference to a then unpublished report by Hooiveld *et al.*, entitled *Second Follow-up of a Dutch Cohort Occupationally Exposed to Phenoxy Herbicides, Chlorophenols, and Contaminants.* Dr Schecter asserted that this unpublished paper provided additional evidence in support of the listing proposal.

The Hooiveld *et al.* report has now been published, and I have had an opportunity to review it in the context of the large body of epidemiologic literature that was examined in depth by the IARC working group during its February 1997 evaluation. On behalf of the American Forest and Paper Association, I have prepared the enclosed written comments on the Hooiveld *et al.* report for consideration by the Subcommittee in its future deliberations regarding the listing proposal for TCDD.

Thank you in advance for your assistance in bringing these comments to the attention of the Subcommittee. If I can provide any further information, please do not hesitate to contact me.

Sincerely,

Raymond S. Greenberg, MD, PhD

Comments on:
Hooiveld *et al:* Second Follow-up of a Dutch Cohort Occupationally Exposed to Phenoxy Herbicides, Chlorophenols, and Contaminants. *Am J Epidemiol* 1998;147:891-901

Raymond S. Greenberg, MD, PhD
Medical University of South Carolina

- **Most of the findings included in this paper were reported previously.**

This paper provides a detailed update of mortality through 1991 for a Dutch cohort of chemical manufacturing workers. A preliminary version of the current paper was published earlier (Hooiveld *et al: Organohalogen Compounds* 1996;30:185-9.) The previous publication of these data was cited in Volume 69 of the International Agency for Research on Cancer (IARC) Monograph on the Evaluation of Carcinogenic Risks to Humans, 1997. That volume provided a comprehensive review of the relationship to cancer risk of exposure to polychlorinated dibenzo-*p*-dioxins. The Dutch cohort was given focused attention in the IARC Monograph (Table 38), because it was one of four industrial populations described in the literature with presumed high levels of exposure.

In addition, the present Dutch cohort was included in the IARC multinational study of cancer risk in relation to occupational exposure to phenoxy herbicides, chlorophenols and dioxins (Kogevinas *et al: Am J Epidemiol* 1997;145:1061-75.) This multinational study also figured prominently in the IARC Monograph (Table 38.) Thus, the recent publication from the Dutch investigators adds relatively little to the already established knowledge base on the carcinogenicity of dibenzo-*p*-dioxins in general, and 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) in particular.

- **The present findings reveal a generalized increase in mortality, rather than associations with specific outcomes.**

When assessing the likelihood that an observed exposure-disease association represents cause and effect, one of the criteria that epidemiologists typically invoke is specificity of the relationship. That is to say, an exposure is more credible as a true cause of an adverse health outcome if it does not appear to be linked indiscriminately to a wide range of outcomes. In Table 4 of the paper, 37 causes of mortality are listed, of which 26 (70%) had possibly suggestive elevations (SMR > 110) in Standardized Mortality Ratios (SMRs), including a variety of non-cancer outcomes. Only five causes of death were lower than expected based upon mortality rates in the general population. While one cannot exclude the possibility that there were true elevations in risk across this diverse set of outcomes, a more plausible explanation is that there was some systematic error (bias) which led to overestimation of the true comparative risk.

- **The present findings are inconsistent with results from other investigations of similar industrial cohorts.**

Another criterion used by epidemiologists to judge purported causal associations is the consistency of findings across studies. To the extent that findings can be independently replicated, greater credence is given to a causal interpretation. In the present context, the results reported by Hooiveld *et al.* are at odds with other key studies in several ways. For example, the excess in overall mortality was not observed in the large cohort compiled by the US National Institute for Occupational Safety and Health (NIOSH), where the SMR was 99 (Fingerhut *et al: New England Journal of Medicine* 1991;324:212-8) or with the large IARC multinational cohort (SMR = 97, Kogevinas *et al.*, 1997). There were suggestions of elevated deaths in the Dutch cohort for diseases affecting mental, nervous, and genitourinary systems, none of which were seen in the IARC multinational cohort. In the Dutch cohort, large elevations in the risk of death from cancers of the bladder and kidney were reported (SMR = 370 and 410 respectively). In contrast, these outcomes were barely above background risk in the IARC multinational cohort (SMRs = 104 and 110, respectively). Similarly, in the NIOSH cohort, the risk of renal cancer was barely above background risk (SMR = 110) and a far more modest elevation was observed in the risk of bladder cancer (SMR = 186) than was reported for the Dutch cohort (SMR = 370).

- **The ability to extrapolate the results of the serum TCDD levels from the surviving workers with measurements to the entire cohort is uncertain.**

The investigators of the Dutch cohort attempted to improve assessment of a possible dose-response relationship by collecting serum TCDD measurements on a subset of the cohort. Back-calculations then were used to impute the peak TCDD levels for these individuals and, based upon employment circumstances, the remainder of the cohort. This is a laudable goal, since many of the prior investigations of the potential adverse effects of TCDD on human health lacked any biological measurements of exposure. Unfortunately, difficulties in obtaining these serum samples, and the consequent low participation rate, limit the utility of this information.

Of the original cohort of almost 1,200 workers, a stratified sampling scheme led to the identification of 144 subjects for serum measurements. Of these individuals, only 47 (33%) actually had useable serum results. The extent to which these individuals validly characterize the original cohort is uncertain and a variety of selection factors may be operating. Those who did participate may have had unusually high exposures, or conversely, may represent the "worried well." In the absence of samples more proximate to the time of exposure and on a larger proportion of the eligible cohort, inferences based upon the serum measurements obtained must be viewed as speculative.

- **In general, the risk of adverse health outcomes does not appear to rise in a smooth graded fashion with increasing imputed maximum levels of TCDD exposure.**

An additional important criterion in judging the likelihood of a causal interpretation is the extent to which the risk of an adverse health outcome rises with increasing level of exposure. The results of the present analysis, (as shown in Table 7 of the current publication), do not show the kind of rising risk from low to medium to high levels of exposure that one would typically associate with causation. For example, the relative risk of all causes of death rises to 1.9 at the medium TCDD level without any further increase at the highest level of exposure. The relative risk for all cancer deaths combined rises to 4.8 at the medium level of exposure, remaining essentially constant for the highest exposure group. The same pattern is seen for cancers of the lung and respiratory system. For malignancies of the urinary organs, the risk rises in the mid-exposure group, only to fall back to the baseline level for the highest exposure category. The elevation in risk for non-Hodgkin's lymphoma essentially was confined to the highest exposure group.

The strongest evidence of a graded increase in risk across exposure levels was seen for accidents, poisoning and violence, with a relative risk rising to 2.2 in the medium exposure group and 5.9 in the highest exposure group. The subset of these events that were classified as suicides also demonstrated an apparent gradient of effect, but this was based on small numbers (two observed deaths each among exposed and non-exposed persons). There is no obvious biological explanation for imputing a cause-and-effect relationship between TCDD exposure and these accidental and violent causes of death.

Although one cannot exclude the possibility of a saturation effect for TCDD exposure by which risk rises at moderate exposure and then levels off or declines, this is not the pattern of risk elevation seen with virtually all known carcinogens. Other epidemiologic studies that have examined TCDD dose (imputed from serum measurements) and risk of cancer deaths have not found the shape of relationship reported by Hooiveld and coworkers. For example, Flesch-Janys *et al.*, (*Am J Epidemiol* 1996;144:716) reported essentially no increase in risk of cancer death up until the highest decile of imputed exposure level. Even at the highest exposure level, the relative risk was half as large as the elevation reported for mid-level exposure by Hooiveld *et al.* Ott and Zober (*Occup Environ Med* 1996;53:606-12) reported a smooth gradient of elevation in risk of death from cancer across four levels of imputed TCDD level of exposure. Again, the highest elevation of relative risk observed by Ott and Zober was less than half that reported by Hooiveld and colleagues in their mid-level of exposure category.

The inconsistency of the Hooiveld *et al.* dose-response data with all other extant epidemiology raises questions about whether the observed risk elevation arose

from some systematic error. That is to say, there may be something about the workers in the exposed groups other than exposure to TCDD *per se* that resulted in their apparent increase in risk for adverse health events. These other risk factors, also known as confounders, might include other industrial exposures encountered by these workers, as well as other non-occupational lifestyle characteristics. The fact that such other exposures might explain the observed relationships was demonstrated in a case-control study nested within the IARC multinational cohort (Kogevinas *et al: Epidemiology* 1995;6:396-402). In that study, the investigators found a graded dose-response relationship between level of TCDD exposure and risk of non-Hodgkin's lymphoma, but they also found an equally strong graded relationship with estimated level of exposure to 2,4,5-trichlorophenoxyacetic acid. In the absence of concurrent data on estimated levels of exposure to other risk factors, one cannot definitively exclude confounding as a possible explanation for the findings of Hooiveld and coworkers.

Conclusion

The findings reported in the present publication have been presented elsewhere in the peer-reviewed literature and were already included in the IARC assessment of the carcinogenicity of TCDD. Although a number of statistical associations between TCDD exposure and adverse effects on human health are reported, the pattern of these findings is not consistent with a causal interpretation. The conventional epidemiologic criteria for causation that are not satisfied by these results are: (1) demonstration of a graded dose-response relationship; (2) consistency with findings from other studies; and (3) specificity of association with particular outcomes.

In conclusion, the present findings add no substantive new evidence regarding possible adverse effects on human health from exposure to TCDD.

MECHANISTIC DATA

Mechanistic Data Are Not Sufficient to Elevate the Classification of Dioxin

The mechanistic data are not a sufficient basis at this time for elevating the classification of dioxin in the *Report on Carcinogens*.

The IARC Working Group concluded that dioxin is carcinogenic to humans by relying on sufficient evidence of carcinogenicity in experimental animals and the presumption that dioxin enhances carcinogenesis through a mechanism involving the aryl hydrocarbon (Ah) receptor, which functions the same way in humans as in experimental animals. Though much is known about Ah receptor binding by dioxin, receptor binding is not sufficient to induce cancer. It is only the first step in a complex sequence of events that are not fully characterized at this time.

Knowledge of dioxin's carcinogenic mechanism in rodents is necessary to establish that it also operates in humans. What is this mechanism? No one knows. Can one point to specific mechanistic steps or events that are known with certainty to contribute to the carcinogenicity of dioxin in rodents? One cannot. While many hypotheses about various aspects of this mechanism have been put forward in the scientific literature, none has been established as scientific fact. Thus, the mechanistic data are not an adequate basis for classification of dioxin as a known human carcinogen.

A. The Mechanism of Dioxin Carcinogenesis Has Not Been Elucidated

Recent mechanistic research has not resolved the many substantial uncertainties surrounding the numerous possible sequelae of Ah receptor binding. Factors and processes other than receptor binding may well provide the critical determinants of whether dioxin is carcinogenic to humans.

1. Enzyme Induction

For example, one of the best-characterized responses to dioxin exposure is induction of drug-metabolizing enzymes such as CYP1A1 via a mechanism of enhanced gene expression which involves binding of dioxin to the cytosolic Ah receptor, transformation and translocation of the occupied receptor to the nucleus, its association with the nuclear protein Arnt, and binding of this complex to dioxin-response elements on DNA that activate gene transcription.

Is this mechanism relevant to the process of carcinogenesis? No one knows. At present, these enzymatic responses serve principally as sensitive biomarkers of dioxin exposure and receptor occupancy.

2. Cell Proliferation

The role of Ah receptor binding in increased cell proliferation, which is widely believed to be a necessary step in carcinogenesis, is also unclear.

- For example, Fox *et al.* (1993) have reported that dioxin-induced changes in cell proliferation occurred in populations of hepatocytes different from those in which there was induction of CYP1A1/CYP1A2.

- More recently, Walker *et al.* (1998) reported that dioxin concentrations in rat liver correlated well with the simple receptor-mediated response of CYP1A1 induction, but did not correlate with the more complex, time-dependent proliferation response.

Walker *et al.* concluded that "The mechanism of TCDD-induced changes in cell proliferation *in vivo* is unknown."

3. Mechanisms Other Than Receptor Binding

Furthermore, mechanisms other than Ah receptor binding and subsequent gene expression have been reported for some of dioxin's effects.

- Enan *et al.* (1998) identified gender-specific dioxin-induced toxicity in guinea pig tissues that occurred far too rapidly to involve gene expression subsequent to the translocation of occupied Ah receptor to the nucleus.

- Puga *et al.* (1992) have reported that dioxin causes a rise in the mRNAs of certain protooncogenes in Hepa-1 cells known to be deficient in the Arnt protein required for DNA binding and subsequent gene transcription.

- After showing that dioxin induces apoptosis in human leukemic T cells that are completely deficient of the Ah receptor, Hossain *et al.* (1998) concluded that "not all the biological effects of TCDD can be explained using this receptor-based model."

Thus, while Ah receptor binding may be a necessary first step in the production of many of dioxin's toxic effects, it appears not to be necessary for all of them. Is Ah receptor binding necessary for carcinogenesis? No one knows. In fact, the 1997 IARC Working Group discussion of this subject concluded that:

> Even though Ah receptor activation is likely to be required for the carcinogenicity of 2,3,7,8-TCDD, its precise role in this process remains unclear.

(IARC Monograph, p. 331)

4. Lack of Specific Mechanism

McGregor *et al.* (1998) summarized the 1997 IARC Working Group's evaluation of possible dioxin-induced carcinogenic mechanisms as follows:

> The mechanistic support for TCDD being a human carcinogen appears limited to its interaction with the AhR (aryl hydrocarbon receptor) and depends on the presumed involvement of this receptor in most, if not all, toxicologic consequences of exposure and the presence of this receptor with similar titrations in all of the animal species tested. However, no specific subsequent steps were proposed to lead to the development of cancers. Instead, it was considered by the Working Group that the plethora of events that could result from AhR interaction --in particular the transcriptional activation of a number of genes --included currently unidentified steps in a carcinogenic pathway.

McGregor, et al., "An IARC Evaluation of Polychlorinated Dibenzo-p-dioxin and Polychlorinated Dibenzofurans as Risk Factors in Human Carcinogenesis," *Environ. Health Persp.* 106:755, 758 (1998)(emphasis added).

In short, we cannot identify with any certainty the specific mechanistic steps or events that contribute to the carcinogenicity of dioxin in rodents. Without knowledge of Dioxin's carcinogenic mechanism in rodents, one cannot confidently conclude that dioxin is known to be carcinogenic in humans. The mechanistic data regarding dioxin carcinogenicity remains insufficient to categorically declare this compound to be a human carcinogen.

B. Mechanistic Experts Believe that the Ah Receptor Binding Evidence is Not Compelling with Regard to Human Carcinogenicity

In October 1997, a group of five scientists internationally renowned for their research on dioxin and carcinogenic mechanisms (Drs. Thomas R. Sutter, Johns Hopkins University, William F. Greenlee and Edward Bresnick, University of Massachusetts Medical Center, Christopher A. Bradfield, University of Wisconsin-Madison, and Alan Poland, CDC/NIOSH), wrote a letter to the *Report on Carcinogens* Subcommittee expressing their strong opposition to the proposal to upgrade dioxin to *Known Human Carcinogen* status in the 9th *Report on Carcinogens.* (Letter attached at Tab B-1.) They, too, called attention to the profound lack of understanding of how dioxin causes any of its toxic effects, an insufficiency in scientific knowledge that continues to the present day.

These eminent scientists state that while evidence of carcinogenicity of dioxin in experimental animals is sufficient, this is not enough to justify classification of dioxin as a known human carcinogen. *"[W]e do not find the additional lines of evidence regarding the Ah receptor and TCDD tissue concentrations at all compelling with regard to the potential human carcinogenicity of TCDD."* (Emphasis added.)

These scientists note that binding with the Ah receptor has been characterized as "necessary but not sufficient" for the production of biochemical and toxic responses to dioxin. The presence and similarity of function of the Ah receptor across species "do not guarantee a similarity of responses, either biochemical or toxic." The presence of the Ah receptor does not predict what, if any, toxic response might occur in a specific tissue in any given species. In fact,

> At the present time, it is impossible to predict what, if any, toxic response might occur in a specific tissue in any given species solely on the basis of Ah receptor presence and functionality. Indeed, a remarkable *disparity* in responses to TCDD exposure exists across tissues and species, yet the mechanisms underlying these marked response differences are multiple and not known or characterized with any degree of scientific certainty (c.f., the review by Schmidt and Bradfield, *Annu Rev Cell Dev Biol* 12:55-89, 1996). Ah receptor occupancy by TCDD is just one, albeit very important, early step in a multiple pathway web of interactions that may, or may not, in any given situation, lead to a toxic response. Lack of understanding of the complex molecular events downstream from receptor occupancy that might or might not culminate in malignancy prevents one from concluding on mechanistic grounds that TCDD is a *Known Human Carcinogen* at the present time."

(Tab B-1, emphasis added.)

As these researchers note, Ah receptor occupancy is just one step in a "web of interactions that may, or may not, in any given situation, lead to a toxic response."

Thus, mechanistic grounds cannot support a listing of dioxin as a known human carcinogen at the present time, and these leaders in mechanistic research have urged NTP to "leave TCDD in its current *Report on Carcinogens* category, namely, *Reasonably Anticipated to be a Human Carcinogen.*"

C. Mechanistic Data Relied on by IARC Consists of Numerous, Varied Unproven Hypotheses

Nowhere in the IARC Working Group's review of potential mechanisms for dioxin carcinogenicity is there an unequivocal statement that a specific mechanism, or even one specific element of such a mechanism, has been identified and then causally linked to tumor production as a result of dioxin exposure. Rather, the hypothesis that Ah receptor binding is a required step in the carcinogenic process is described by the Working Group only as likely, and "its precise role in this process remains unclear." (Monograph at 331, emphasis added.)

A close review of section 4.6 of the Monograph shows just how many hypotheses regarding dioxin's carcinogenic mechanism(s) have been proposed. None has been confirmed as the actual mechanism responsible for any of the increases in tumor incidence that have been observed following dioxin exposure.

The IARC Working Group was unable to identify specific events in a carcinogenic pathway common to all affected species that were by accelerated or amplified by dioxin exposure. Reference is made to the transcriptional activation of a number of genes, but such activation by dioxin has not been causally linked to the induction of tumors in any species. Indeed, the conclusions apparent from the discussion of carcinogenic mechanisms in section 4.6 of the IARC Monograph are that:

1) binding to the Ah receptor is not sufficient to cause cancer or any other form of toxicity;

2) events beyond receptor binding that lead ultimately and unequivocally to tumor formation by dioxin have yet to be identified; and

3) causal linkages of such events to receptor occupancy by dioxin have yet to be firmly established by scientific experimentation.

The inescapable conclusion from the Working Group's evaluation of potential carcinogenic mechanisms of dioxin is that this body of information is comprised of numerous and varied plausible hypotheses, none of which has been demonstrated as scientific fact. These hypotheses regarding the potential mechanism of carcinogenicity of dioxin cannot overcome the severe limitations of the human evidence.

Current scientific evidence does not justify upgrading 2,3,7,8-tetrachloro-dibenzo-*para*-dioxin to *Known Human Carcinogen* status in the 9th *Report on Carcinogens*. Dioxin should continue to be classified as *Reasonably Anticipated to be a Human Carcinogen*.

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Via Facsimile: 919/541-0295

28 October 1997

NTP Board of Scientific Counselors' *Report on Carcinogens* Subcommittee
c/o Dr. Larry G. Hart, Executive Secretary
National Institute of Environmental Health Sciences
Research Triangle Park NC

Dear Subcommittee Members:

We the undersigned believe the proposal to upgrade 2,3,7,8-tetrachlorodibenzo-*para*-dioxin (TCDD) to *Known Human Carcinogen* status in the 9th *Report on Carcinogens* is not supported by the scientific evidence. TCDD should be left in its present classification category, *Reasonably Anticipated to be a Human Carcinogen.*

In February 1997, an IARC Working Group summarized its evaluation of the scientific evidence regarding the carcinogenic risks posed by TCDD as follows:

> "There is *limited evidence* in humans for the carcinogenicity of 2,3,7,8-tetrachloro-dibenzo-*para*-dioxin."
>
> "There is *sufficient evidence* in experimental animals for the carcinogenicity of 2,3,7,8-tetrachlorodibenzo-*para*-dioxin."

The IARC Monographs Preamble describes *limited evidence* in humans as follows:

> "A positive association has been observed between exposure to the agent, mixture or exposure circumstance and cancer for which a causal interprestion is considered by the Working Group to be credible, but chance, bias or confounding could not be ruled out with reasonable confidence."

In comparison, the *Report on Carcinogens* criterion for listing a substance as a *Known Human Carcinogen* is:

> "There is sufficient evidence of carcinogenicity from studies in humans which indicates a causal relationship between exposure to the agent, substance or mixture and human cancer."

Presuming the accuracy of the IARC Working Group's evaluation, the evidence for the carcinogenicity of TCDD in humans falls short of satisfying this criterion. We are aware of no evidence, be it epidemiologic, toxicologic, or mechanistic, that would justify upgrading the *Report on Carcinogens* classification of TCDD to a *Known Human Carcinogen* at the present time.

We realize that the IARC Working Group placed TCDD in IARC's Group 1 (carcinogenic to humans) based on 1) the sufficiency of evidence for the carcinogenicity of TCDD in experimental animals, 2) the presence and similarity of function of the evolutionarily conserved Ah receptor in humans and experimental animals, and 3) the similarity of TCDD tissue concentrations in heavily exposed human populations in which an increased overall cancer risk was observed to those in rats exposed to carcinogenic doses in bioassays.

We agree with the Working Group's assessment that the evidence for the carcinogenicity of TCDD in experimental animals is sufficient. However, this evidence is not sufficient to classify TCDD as a *Known Human Carcinogen*, and we do not find the additional lines of evidence regarding the Ah receptor and TCDD tissue concentrations at all compelling with regard to the potential human carcinogenicity of TCDD.

Consider first the scientific evidence regarding the Ah receptor. Its role in the production of biochemical and toxic responses to TCDD exposure has been characterized fairly as *necessary but not sufficient.* Thus, its presence and similarity of function across species do not guarantee a similarity of responses, either biochemical or toxic. At the present time, it is impossible to predict what, if any, toxic response might occur in a specific tissue in any given species solely on the basis of Ah receptor presence and functionality. Indeed, a remarkable *disparity* in responses to TCDD exposure exists across tissues and species, yet the mechanisms underlying these marked response differences are multiple, and not known or characterized with any degree of scientific certainty (c.f., the review by Schmidt and Bradfield, *Annu Rev Cell Dev Biol* 12:55-89, 1996). Ah receptor occupancy by TCDD is just one, albeit very important, early step in a multiple pathway web of interactions that may, or may not, in any given situation, lead to a toxic response. Lack of understanding of the complex molecular events downstream from receptor occupancy that might or might not culminate in malignancy prevents one from concluding on mechanistic grounds that TCDD is a *known human carcinogen* at the present time.

Next, consider the evidence regarding the similarity of TCDD tissue concentrations (i.e., esitmated body burdens) in heavily exposed human populations to those in rats exposed to carcinogenic TCDD doses. While elevated cancer risks have been reported in studies of some heavily exposed human populations, the IARC Working Group was unable to confidently rule out chance, bias or confounding as being responsible for them. This means that TCDD exposure might not have been the cause of the reported excess risks. If the excess risks were in fact due to factors other than heavy TCDD exposure, then this evidence shows only that certain heavily exposed human populations developed TCDD body burdens similar to those in rats that received carcinogenic bioassay doses. It shows nothing about the actual carcinogenicity of TCDD in humans.

In summary, the scientific evidence is not sufficient at present to conclude that TCDD is a *Known Human Carcinogen.* We encourage the Subcommittee to leave TCDD in its current *Report on Carcinogens* category, namely, *Reasonably Anticipated to be a Human Carcinogen.*

Sincerely,

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